OBSERVATIONS AND RESULTS

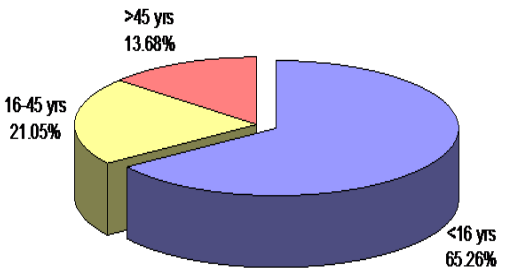
Our study evaluated the peripheral blood and bone marrow aspirate smears in acute myeloid leukemia for morphological diagnosis and compared it with their cytochemical diagnosis and immunophenotypic profile. We collected data from 95 patients, of all ages and either sex.

A. Patient characteristics:

The basic characteristics of the patients are presented in Tables 1-5 and also shown graphically alongside.

Table 1: Distribution of Study Population according to age (N=95)

Age Group	Number	Percentage
Children (<16 yrs)	62	65.26
Adults (>16 yrs)	33	34.74
Young adults (16-45 yrs)	20	21.05
Old adults (>45 yrs)	13	13.68

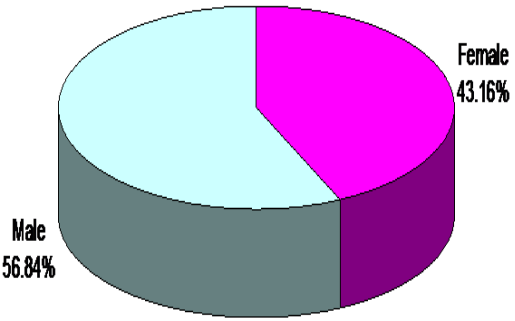


The present study consisted of 95 patients, out of these majority were children <16 years (65.26%), 20 (21.05%) were aged 16-45 years and 13 (13.68%) were aged >45 years. We know that AML is more common in adults, contrary to our findings here. Explaination to this finding lies in the fact that in our institution, there is more admission of pediatric patients.

Also these findings came out to be statistically insignificant.

Table 2: Distribution of Study Population according to Gender (N=95)

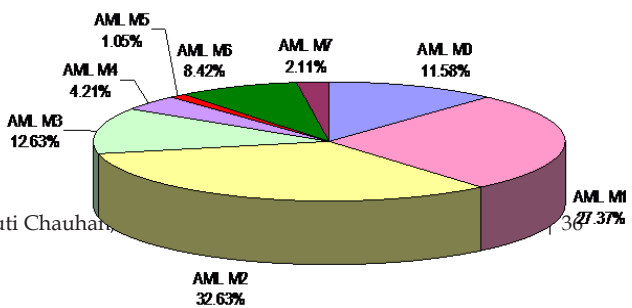
Gender	Number	Percentage
Female	41	43.16
Male	54	56.84



Prevalence was found to be higher in males (56.84%)as compared to females (43.16%). Male:Female ratio was 1.3:1.(TABLE 2).

Table 3a: Distribution of Study Population according to Morphological Diagnosis (N=95)

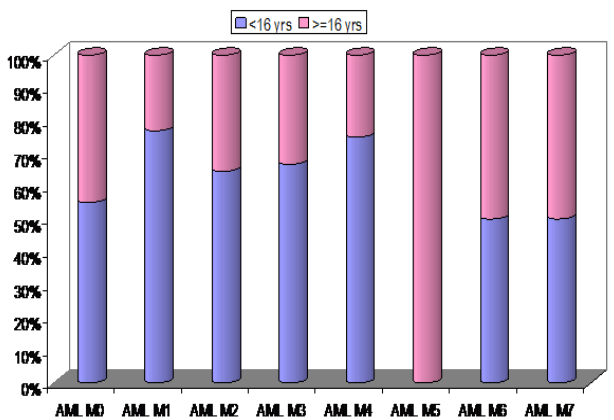
Morphological Diagnosis	Number	Percentage
AML M0	11	11.58
AML M1	26	27.37
AML M2	31	32.63
AML M3	12	12.63
AML M4	4	4.21
AML M5	1	1.05
AML M6	8	8.42
AML M7	2	2.11



Most common morphological diagnosis was AML-M2 (32.63%) followed by AML-M1 (27.37%), AML-M3 (12.63%), AML-M0 (11.58%), AML-M6 (8.42%), AML-M4 (4.21%), AML-M7 (2.11%) and least common was AML-M5 (1.05%).

Table 3(b): Comparison of morphological diagnosis in Children and adults

Morphological Diagnosis	Total (N=95)		Age <16 yrs (n=62) Children		Age >16 (n=33) Adults	Statistical Significance	
	No.	%	No.	%	χ^2	P	
AML M0	11	6	55.54	5	45.45	5.206 (df=7)	0.635
AML M1	26	20	76.92	6	23.08		
AML M2	31	20	64.52	11	35.48		
AML M3	12	8	66.67	4	33.33		
AML M4	4	3	75.00	1	25.00		
AML M5	1	0	0.00	1	100.00		
AML M6	8	4	50.00	4	50.00		
AML M7	2	1	50.00	1	50.00		



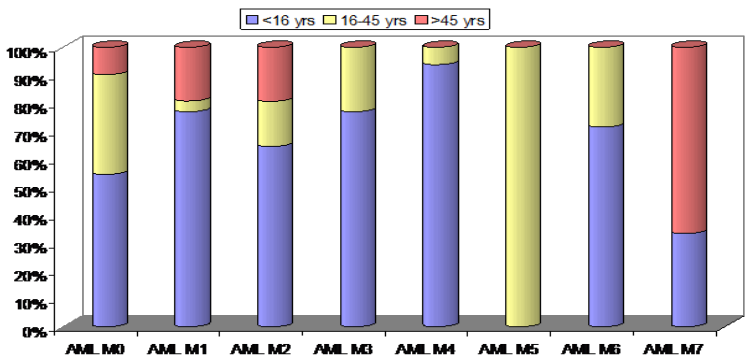
Though proportion of subjects aged <16 years was high in AML-M1 (76.92%) and AML-M4 (75.00%) as compared to subjects with other morphological

diagnosis but this difference was not found to be statistically significant (p=0.635).As per the table, maximum number of cases were from AML M1 and M2, common subtype in the pediatric population while AML M5 is the least common. Also, AML M2 had maximum number of cases in the adult population while AML M4,5 and 7 are the less common subtypes.

Table 3(c): Comparison of morphological diagnosis in different age groups

Morphological Diagnosis	Total (N=95)		Age <16 yrs (n=62)		Age 16-45 yrs (n=20)		>45 yrs (n=13)	
	No.	%	No.	%	No.	%		
AML M0	11	6	55.54	4	36.36	1	9.99	
AML M1	26	20	76.92	1	3.85			
5	19.23							
AML M2	31	20	64.52	5	16.13	6	19.35	
AML M3	12	8	66.67	4	20.00	0	0.00	
AML M4	4	3	75.00	1	5.00	0	0.00	
AML M5	1	0	0.00	1	5.00	0	0.00	
AML M6	8	4	50.00	4	20.00	0	0.00	
AML M7	2	1	50.00	0	0.00	1	100.00	

21.334=^{2} (df=14); p=0.093 (Non-Significant)

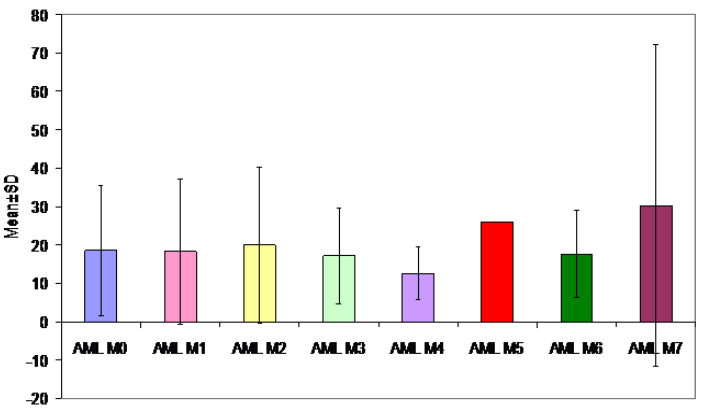


Majority of patients of all the morphological diagnosis were <16 years of age except the only patient diagnosed as AML-M5 (n=1) who was aged between 16-45 years. Difference in age of patients with different morphological diagnosis was not found to be statistically significant (p=0.093).In the age group >45 years, the proportion was highest in AML M7 (100%) but maximum number of cases were seen in AML M2.

Table 3(d): Comparison of Age in different morphological diagnoses

Morphological Diagnosis	No. of subjects	Min.	Max.	Mean	S.D.
AML M0	11	1.50	47.00	18.59	16.89
AML M1	26	1.50	65.00	18.29	18.96
AML M2	31	1.50	80.00	19.97	20.24
AML M3	12	3.00	42.00	17.17	12.47
AML M4	4	7.00	22.00	12.50	6.86
AML M5	1	26.00	26.00	26.00	.
AML M6	8	2.00	35.00	17.63	11.33
AML M7	2	0.58	60.00	30.29	42.01
Total	95	0.58	80.00	18.76	17.63

F=0.246; p=0.972



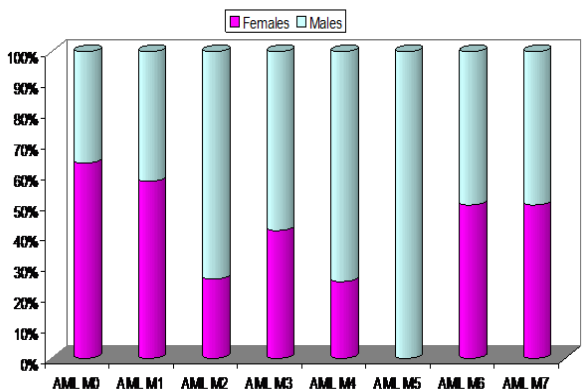
Age of patients ranged from 7 months to 80 years with a mean age of 18.76 and standard deviation of 17.63 years. Table 3d shows that the minimum age was seen in AML M7 while the maximum was seen in M2 subtype. The highest mean is seen in the AML M5 subtype.

Age of patients of AML-M7 (30.29±42.01 years) and AML-M5 (26.00 years; n=1) was found to be higher than that having other morphological diagnosis. Minimum mean age was found to be in AML-M4 patients.

Difference in mean age of patients with different diagnosis was not found to be statistically significant (p=0.972).

Table 4: Comparison of morphological diagnosis in Females and Males

Morphological Diagnosis	Total (N=95)		Female (n=41)		Male (n=54)	
Statistical Significance						
	No.	%	No.	%	^{2}	p
AML M0 11	7	63.64	4	36.36	9.422 (df=7)	0.224 (NS)
AML M1 26	15	57.69	11	42.31		
AML M2 31	8	25.81	23	74.19		
AML M3 12	5	41.67	7	58.33		
AML M4 4	1	25.00	3	75.00		
AML M5 1	0	0.00	1	100.00		
AML M6 8	4	50.00	4	50.00		
AML M7 2	1	50.00	1	50.00		

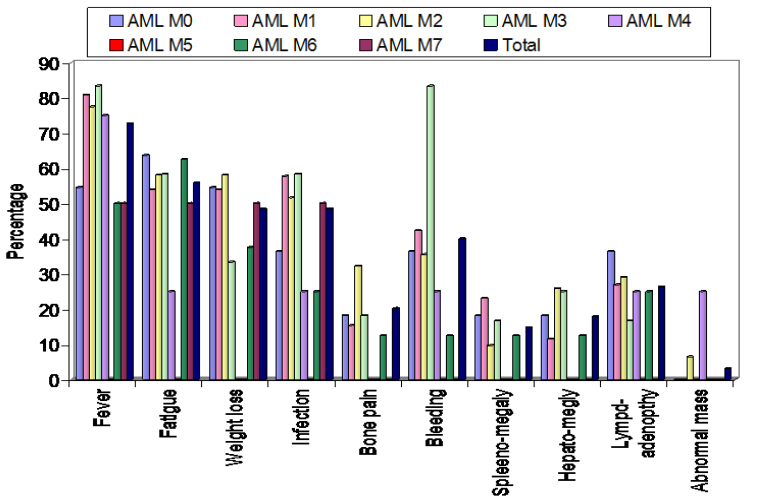


Majority of patients of AML-M0 and AML-M1 were females while majority of patients diagnosed as AML-M2, AML-M3 and AML-M4 were males. There was equal gender based distribution in AML M6 and M7. Only patient diagnosed as AML-M5 was male. Despite proportional difference in gender of subjects with different morphological diagnosis the difference was not found to be statistically significant (p=0.224).

Table 5: Comparison of Presenting Symptoms and signs in different morphological diagnoses

	M0 (n=11)		M1 (n=26)		M2 (n=31)		M3 (n=12)		M 4
(n=4)	M5 (n=1)	M6 (n=8)	M7 (n=2)	Total (N=95)		^{2}	p		
Fever	6	21	24	10	3	0	4	1	69
8.967	0.255								
	54.55	80.77	77.42	83.33	75.00	0.00	50.00	50.00	
72.63									
Fatigue	7	14	18	7	1	0	5	1	53
3.383	0.847								
	63.64	53.85	58.06	58.33	25.00	0.00	62.50	50.00	
55.79									
Weight loss		6	14	18	4	0	0	3	1
46	7.798	0.351							
	54.55	53.85	58.06	33.33	0.00	0.00	37.50	50.00	
48.42									
Infection	4	15	16	7	1	0	2	1	46
5.710	0.574								
	36.36	57.69	51.61	58.33	25.00	0.00	25.00	50.00	
48.42									
Bone pain	2	4	10	2	0	0	1	0	19
5.289	0.625								
	18.18	15.38	32.26	18.18	0.00	0.00	12.50	0.00	
20.21									
Bleeding	4	11	11	10	1	0	1	0	38
14.666	0.041								
	36.36	42.31	35.48	83.33	25.00	0.00	12.50	0.00	
40.00									

Splenomegaly		2	6	3	2	0	0	1	0
14	3.452	0.840							
	18.18	23.08	9.68	16.67	0.00	0.00	12.50	0.00	
14.74									
Hepatomegaly		2	3	8	3	0	0	1	0
17	4.133	0.764							
	18.18	11.54	25.81	25.00	0.00	0.00	12.50	0.00	
17.89									
Lymphadenopathy		4	7	9	2	1	0	2	0
25	2.354	0.938							
	36.36	26.92	29.03	16.67	25.00	0.00	25.00	0.00	
26.32									
Abnormal mass									
(orbital swelling)		0	0	2	0	1	0	0	0
3	9.301	0.232							
	0.00	0.00	6.45						
0.00	25.0	0.00	0.00	0.00	3.16				



Overall most common symptom was fever (72.63%) followed by Fatigue (55.79%), weight loss (48.42%) and infection (48.42%), the less common ones were bleeding, lymphadenopathy, bone pain, hepatomegaly and splenomegaly while the least common symptom was abnormal mass

(3.16%).

Fever was present in 69 (72.63%) subjects. Proportion of patients presenting with fever was higher in AML M3 (83.33%), AML M1 (80.77%), AML M2 (77.42%) and AML M4 (75.00%) as compared to patients with other morphological diagnosis (0 to 54.55%). Difference in fever as presenting symptom in different morphological diagnosis was not found to be statistically significant (p=0.255).

Fatigue was present in 53 (55.79%) subjects. Proportion of patients presenting with fatigue was higher in AML M0 (63.64%) AML M6 (62.50%) and AML-M2 (58.06%) as compared to patients with other morphological diagnosis (0 to 55.79%). Difference in fatigue as presenting symptom in different morphological diagnosis was not found to be statistically significant (p=0.847).

Weight loss was present in 46 (48.42%) subjects. Proportion of patients presenting with weight loss was higher in AML M2 (58.06%), AML M0 (54.55%), AML M1 (53.85%) as compared to patients with other

Diagnosis. However, this was not found to be statistically significant (p=0.574).

Bone pain was found in 19 (20.21%). Proportional differences in bone pain as presenting symptoms in different morphological diagnosis was not found to be statistically significant (p=0.625).

Bleeding was found in 38 (40.00%) patients. Proportion of patients diagnosed as AML M3 (83.33%) was found to be significantly higher than other morphological diagnosis (p=0.041%). This was followed by AML M1 and M0 and least in AML M5 and M7 types.

Splenomegaly was found in 14 (14.74%) patients. Proportional differences in splenomegaly as presenting symptoms in different morphological diagnosis was not found to be statistically significant (p=0.840). It was most common in AML M1 subtype followed by AML M0 and least common in AML M4, M5 and M7.

Hepatomegaly was found in 17 (17.89%) patients. Proportional differences in hepatomegaly as presenting symptoms in different morphological diagnosis was not found to be statistically

significant (p=0.764)

It was most common in AML M2 and M3 subtype and least common in AML M4 and M5.

Lymphadenopathy was found in 25 (26.32%) patients. Proportional differences in Lymphadenopathy as presenting symptoms in different morphological diagnosis was not found to be statistically significant (p=0.938).

It was most commonin AML M0 subtype followed by AML M2 and M1 and least common in AML M5 and M7.

Abnormal mass (i.e. orbital swelling) was found in only 3 (3.16%) patients. Abnormal mass was observed in 2 (6.45%) patients of AML-M2 and 1 (25.0%) patients of AML-M4. Proportional differences in abnormal mass as presenting symptoms in above two morphological diagnosis was not found to be statistically significant (p=0.232).

B. Morphological and cytochemical studies:

Table 6a: Comparison of Hematological Variables in different morphological diagnoses

Variable	Morphological Diagnosis	No. of subjects	Min.	Max.	Mean	S.D.	F	p
PBS-Hb	AML M0	11	3.90	11.00	6.80	2.47	0.853	0.547
	AML M1	26	3.00	9.00	5.70	1.56		
	AML M2	31	2.60	12.60	6.14	2.21		
	AML M3	12	4.10	10.30	6.18	1.69		
	AML M4	4	4.40	5.10	4.73	0.33		
	AML M5	1	4.00	4.00	4.00	.		
	AML M6	8	5.30	9.10	6.23	1.30		
	AML M7	2	5.60	7.10	6.35	1.06		
	Total	95	2.60	12.60	6.03	1.89		
TLC	AML M0	11	3300	410000	68318	118885	0.963	0.464
	AML M1	26	1200	160000	31242	40005		
	AML M2	31	1100	230000	45655	55668		
	AML M3	12	3500	112000	22975	31304		

	AML M4	**4**	**28000**	**81600**	**46300**	**24355**		
	AML M5	**1**	**11600**	**11600**	**11600**	**.**		
	AML M6	**8**	**1800**	**22400**	**14525**	**8460**		
	AML M7	**2**	**9000**	**21000**	**15000**	**8485**		
	Total	**95**	**1100**	**410000**	**37872**	**57400**		
PC	**AML M0**	**11**	**10000**	**78000**	**26182**	**18324**	**0.699**	**0.673**
	AML M1	**26**	**8000**	**170000**	**42150**	**39697**		
	AML M2	**31**	**8000**	**245000**	**44116**	**54238**		
	AML M3	**12**	**10000**	**48000**	**23450**	**11276**		
	AML M4	**4**	**10000**	**55000**	**35250**	**23243**		
	AML M5	**1**	**35000**	**35000**	**35000**	**.**		
	AML M6	**8**	**20000**	**160000**	**53625**	**50746**		
	AML M7	**2**	**18200**	**110000**	**64100**	**64912**		
	Total	**95**	**8000**	**245000**	**39643**	**41903**		

The haemoglobin levels ranged from 2.6 to 12.6 gm% with a mean value of 6.03 gm% ±1.89(SD). The lowest as well as the highest value was found in AML M2 subtype. AML M5 had the lowest mean value while AML M0 had the highest mean value.

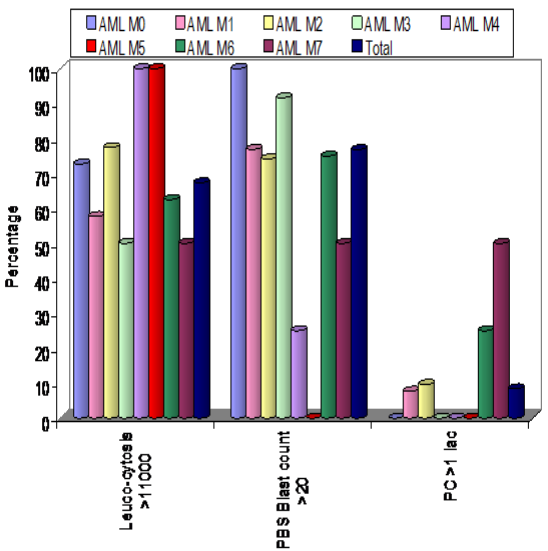
The hemoglobin levels were found to be <10 gm/dl in 91 (95.79%) patients while in 4 patients (4.21%) hemoglobin levels were found to be >10 gm/dl. Out of these 4 patients, 1 was diagnosed as AML-M0, 2 as AML M2 and 1 as AML-M3.

The total leukocyte count ranged from 1100 cells/cu.mm to 4,10,000 cells/cu.mm.The mean value was 37,872 cell/cu.mm ±57400 cells/cu.mm(SD). The lowest value was seen in AML M2 while the highest value was found in AML M0 subtype.AML M5 had the lowest mean value while the AML M0 had the highest mean value.

The platelet count ranged from 1100 /cu.mm to 4,10,000 / cu.mm.The mean value was 37,872 /cu.mm ±57400 /cu.mm(SD). The lowest value was seen in AML M2 while the highest value was found in AML M0 subtype.AML M5 had the lowest mean value while the AML M0 had the highest mean value.

Table 6b: Comparison of Hematological Variables in different morphological diagnoses

	M0 (n=11)		M1 (n=26)		M2 (n=31)		M3 (n=12)		M 4
(n=4)	M5 (n=1)	M6 (n=8)	M7 (n=2)	Total (N=95)		^{2}	p		
Leucocytosis >11000cells/cumm			8	15	24	6	4	1	
5	1	64	7.105	0.418					
	72.73	57.69	77.42	50.00	100.00	100.00	62.50	50.00	67.37
PBS Blast count >20%			11	20	23	11	1	0	6
1	73	15.104	0.035						
	100.00	76.92	74.19	91.67	25.00	0.00	75.00	50.00	76.84
PC >1 lac/cumm	0	2	3	0	0	0	2	1	
8	9.991	0.189							
	0.00	7.69	9.68	0.00	0.00	0.00	25.00	50.00	8.42



Leucocytosis (>11,000 cells/ cu.mm) was found in 64 (67.37%) patients. Proportion of patients with leucocytosis was found in higher proportion of patients of AML-M4 (100.0%), AML-M5 (n=1; 100.0%), AML-M2 (77.42%) and

AML-M0 (72.73%) as compared to patients with other morphological diagnosis but this difference was not found to be statistically significant (p=0.418).

Leucocytopenia (<11,000/ cu.mm) was found in 31 (32.63%) patients of the study population. Proportion of patients with leucocytopenia was found in higher proportion of patients of AML-M3 (50.0%), AML-M7 (50.0%) and AML-M1 (42.31%) as compared to patients with other morphological diagnosis.

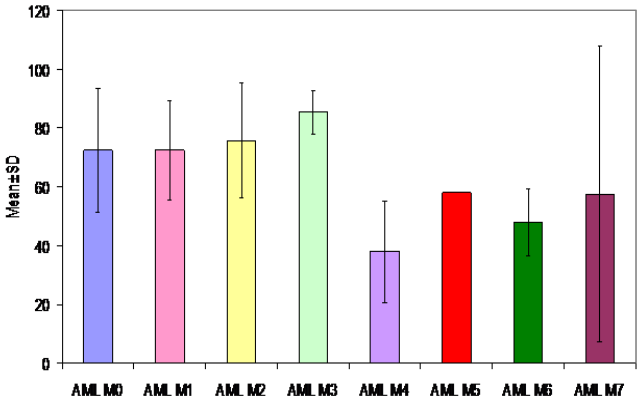
PBS-blast count (>20%) was found in 73 (76.84%) patients. Proportion of patients with PBS-blast count >20% was highest in AML M0 and AML M3 and significantly lower in AML-M4 (25.0%) as compared to patients of other morphological diagnosis. The single patient diagnosed as AML-M5 had PBS-blast count <20% (ie was in aleukemic phase).

Platelet count >1 lac were found in only 8 (8.42%) patients. None of the patients in AML M0, M3, M4, and M5 had platelet count more than 1 lac. Thus, all the cases in these subtypes showed thrombocytopenia. Though proportion of patients with platelet count >1 lac was higher in AML-M7 (50.0%) and AML-M6 (25.00%) as compared to patients with other morphological diagnosis but this difference was not found to be statistically significant.

Table 7: Comparison of Bone Marrow Blast Cells in different morphological diagnoses

Morphological Diagnosis	No. of subjects	Min.	Max.	Mean	S.D.
AML M0	11	23.00	93.00	72.45	21.14
AML M1	26	37.00	95.00	72.42	16.76
AML M2	31	28.00	98.00	75.77	19.43
AML M3	12	71.00	97.00	85.33	7.27
AML M4	4	21.00	62.00	38.00	17.26
AML M5	1	58.00	58.00	58.00	.
AML M6	8	23.00	61.00	48.00	11.36
AML M7	2	22.00	93.00	57.50	50.20
Total	95	21.00	98.00	71.18	20.65

F=5.547; p<0.001



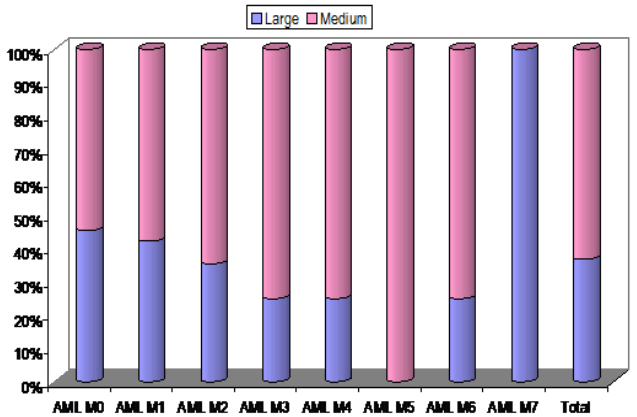
Number of bone marrow blast cells were found to be significantly lower in patients diagnosed as AML M4 (38.00+17.26), AML M5 (58.00) and AML M6 (57.50+50.20) as compared to patients with other diagnosis. None of the patient had blast count <20%.

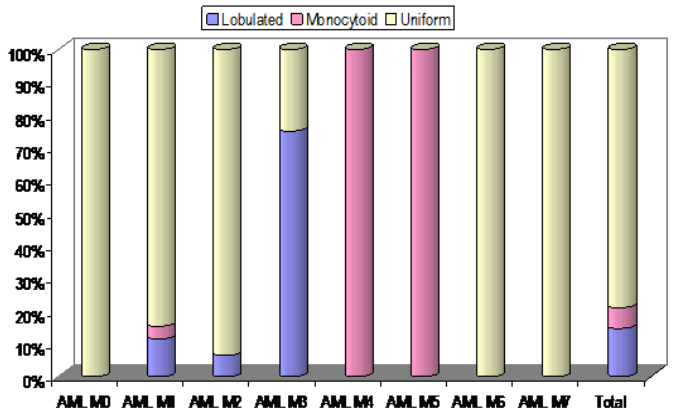
Table 8: Comparison of Morphology of Blasts in different morphological diagnoses

	M0 (n=11)	M1 (n=26)	M2 (n=31)	M3 (n=12)	M4 (n=4)	M5 (n=1)	M6 (n=8)	M7 (n=2)	Total (N=95)	χ^2	P
Size											
Large	5	11	11	3	1	0	2	2	35	6.167	0.520
	45.45	42.31	35.48	25.00	25.00	0.00	25.00	100.00	36.84		
Medium	6	15	20	9	3	1	6	0	60		
	54.55	57.69	64.52	75.00	75.00	100.00	75.00	0.00	63.16		
Shape											
Lobulated		0	3	2	9	0	0	0	14	119.297	<0.001
	0.00	11.54	6.45	75.00	0.00	0.00	0.00	0.00			

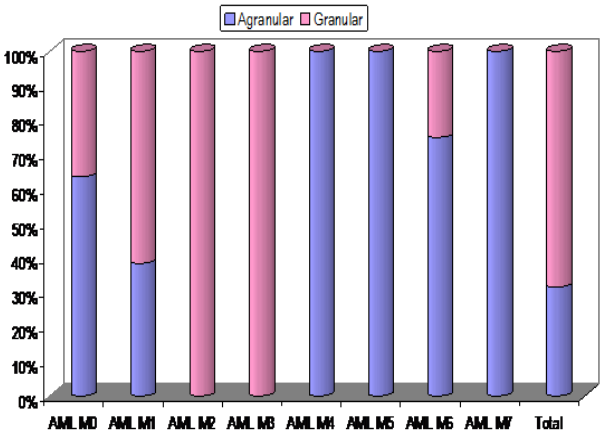
14.74									
Monocytoid		0	1	0	0	4	1	0	0
6									
	0.00	3.85	0.00	0.00	100.00	100.00	0.00	0.00	6.32
Uniform	11	22	29	3	0	0	8	2	75
	100.00	84.62	93.55	25.00	0.00	0.00	100.00	100.00	78.95
Cytoplasm									
Agranular		7	10	0	0	4	1	6	2
30	47.796	<0.001							
	63.64	38.46	0.00	0.00	100.00	100.00	75.00	100.00	31.58
Granular	4	16	31	12	0	0	2	0	65
	36.36	61.54	100.00	100.00	0.00	0.00	25.00	0.00	68.42
Dysplasia									
Dysplasia	0	2	6	1	0	0	0	0	9	6.365
0.498									
	0.00	7.69	19.35	8.33	0.00	0.00	0.00	0.00	9.47
No dysplasia	11	24	25	11	4	1	8	2	86
	100.00	92.31	80.65	91.67	100.00	100.00	100.00	100.00	91.53

Out of 95 patients included in the study in majority cases (n=60; 63.16%) , blast size was found to be medium. All the patients diagnosed as AML-M7 had larger blast size as compared to other cases. Despite proportional difference in size of blasts in patients with different morphological diagnoses, this difference was not found to be statistically significant (p=0.520).

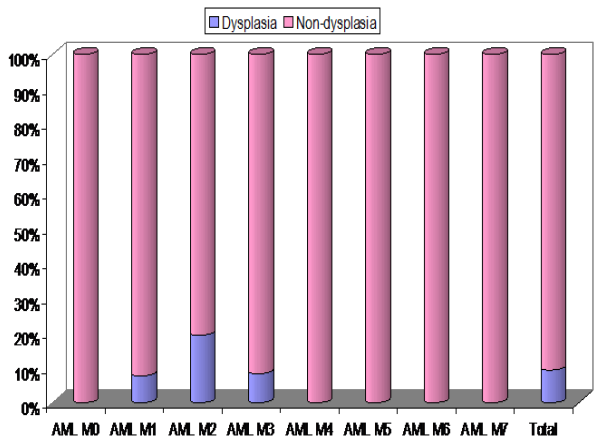




Shape of blasts in majority of subjects in our study was uniform (n=75; 78.95%). All the subjects diagnosed as AML-M4 and AML-M5 had predominantly monocytoid shape. Lobulated shape was found in most of the patients (9 ;75%) diagnosed as AML-M3. Difference in shapes of blasts in patients with different morphological diagnosis was found to be statistically significant ($p<0.001$).



Cytoplasm was found to be granular in majority of patients of our study (n=65; 68.42%). All the patients diagnosed as AML-M2, AML-M3 had granular cytoplasm while the patients of AML-M4, AML-5 and AML-M7 had agranular cytoplasm. Difference in granularity of cytoplasm in patients with different morphological diagnosis was found to be statistically significant ($p<0.001$).

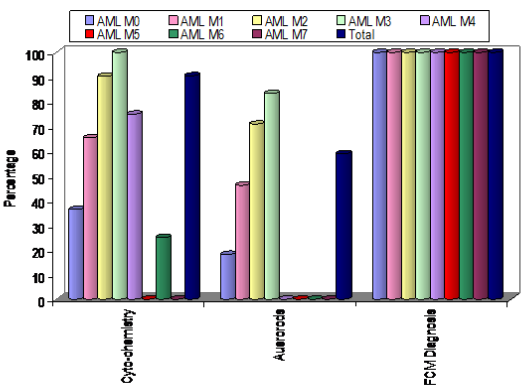


Dysplasia was found in 9 (9.47%) patients. None of the patients diagnosed as AML-M0, AML-M4, AML-M5, AML-M6 and AML-M7 had dysplasia. Dysplasia was found in higher proportion of patients diagnosed as AML-M2 (19.35%) as compared to

AML-M3 (8.33% and AML-M1 (7.69%).These cases of AML may have been the ones that had progressed from myelodysplastic syndrome. Difference in dysplasia in patients with different morphological diagnosis was not found to be statistically significant (p=0.498).

Table 9: Comparison of Positive Cytochemistry and Auer Rods in different morphological diagnoses

	M0 (n=11)	M1 (n=26)	M2 (n=31)	M3 (n=12)	M4 (n=4)	M5 (n=1)	M6 (n=8)	M7 (n=2)	Total	p
Cyto-chemistry	4	17	28	12	3	0	2	0	66/95	31.864	<0.001
6.36	65.38	90.32	100.00	75.00	0.00	25.00	0.00	69.47			
	4	17	28	12	3				64/84		
76.19%											
Auer rods	2	12	22	10	0	0	0	0	46	30.329	<0.001
	18.18	46.15	70.97	83.33	0.00	0.00	0.00	0.00	48.42		
46/84											
54.74%											
FCM Diagnosis	11	26	31	12	4	1	8	2	95		
	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00		



The cytochemical staining that was done using Myeloperoxidase (MPO) method was found to be positive in 64/84 cases (76.19%), excluding the AML M5, M6 and M7 cases.Maximum positivity was seen in AML M3(100%) category, followed by AML M2(90.32%). Statistically, the positivity of MPO in identifying the myeloid lineage was found to be significant. ($p<0.001$)

Auer rods were found to be positive in 46/84 cases excluding the AML M5, M6 and M7 subtypes (54.74%). Maximum positivity was seen in AML M3(83.3%) category, followed by AML M2(70.97%) , M1 (46.12%) and M0 (18.18%) while not seen in any case of AML M4,M5,M6 and M7(0%).

Table10: Immune markers in different morphological diagnoses (mean±SD)

Variable	Morphological Diagnosis	No. of subjects		Min.	Max.	Mean	S.D.	F	P
CD13	AML M0	11	0.00	94.00	69.55	32.74	0.514	0.796	
	AML M1	24	0.00	96.00	62.15	30.11			
	AML M2	27	7.00	99.00	69.79	27.74			
	AML M3	10	0.00	98.00	75.49	29.71			
	AML M4	4	65.00	93.00	81.70	11.86			
	AML M6	6	43.00	98.00	62.05	23.84			
	AML M7	1	58.40	58.40	58.40	.			
	Total	83	0.00	99.00	68.11	28.23			
CD33	AML M0	11	8.00	97.00	73.34	25.54	1.860	0.099	
	AML M1	24	7.00	99.00	75.55	26.43			
	AML M2	27	0.00	99.00	75.33	25.87			
	AML M3	10	83.00	99.00	94.79	4.63			
	AML M4	4	45.00	80.00	64.25	15.00			
	AML M6	6	85.00	98.30	95.72	5.27			
	AML M7	1	80.90	80.90	80.90	.			
	Total	83	0.00	99.00	78.48	23.95			
HLADR	AML M0	5	0.00	95.00	73.96	41.42	4.603	0.001 (Sig)	
	AML M1	11	0.00	92.00	66.55	25.66			
	AML M2	17	0.00	99.60	59.78	31.88			
	AML M3	10	0.00	54.00	9.90	20.98			
	AML M4	4	0.00	69.00	44.50	30.47			
	AML M6	5	0.00	81.20	62.20	34.84			
	AML M7	1	76.00	76.00	76.00	.			
	Total	53	0.00	99.60	52.49	35.66			

MPO	AML M0	8	0.00	83.00	36.88	32.45	4.137	0.002
	AML M1	18	0.00	89.00	48.22	28.33		
	AML M2	17	0.00	94.00	61.51	25.26		
	AML M3	8	31.00	99.30	74.41	20.57		
	AML M4	3	0.00	66.00	32.67	33.01		
	AML M6	6	0.00	85.10	14.18	34.74		
	AML M7	1	0.00	0.00	0.00	.		
	Total	61	0.00	99.30	48.97	32.12		
CD3	AML M0	5	0.00	1.00	0.24	0.43	1.404	0.257
	AML M1	12	0.00	7.00	1.44	2.11		
	AML M2	11	0.00	2.00	0.30	0.64		
	AML M3	5	0.10	2.00	0.54	0.82		
	AML M4	2	0.00	0.00	0.00	0.00		
	AML M6	0	.	.	.	.		
	AML M7	0	.	.	.	.		
	Total	35	0.00	7.00	0.70	1.40		
CD2	AML M0	10	0.00	34.00	3.42	10.74	0.531	0.752
	AML M1	16	0.00	97.00	8.75	25.46		
	AML M2	21	0.00	32.00	1.62	6.97		
	AML M3	4	0.00	46.00	11.50	23.00		
	AML M4	2	0.00	4.00	2.00	2.83		
	AML M6	2	0.00	0.00	0.00	0.00		
	AML M7	0	.	.	.	.		
	Total	55	0.00	97.00	4.69	16.13		
CD7	AML M0	11	0.00	78.00	15.27	27.37	1.035	0.410
	AML M1	22	0.00	85.00	21.35	31.76		
	AML M2	25	0.00	65.10	4.55	14.16		
	AML M3	9	0.00	94.70	10.71	31.50		
	AML M4	3	0.00	0.10	0.03	0.06		
	AML M6	6	0.00	65.00	11.13	26.39		
	AML M7	1	1.60	1.60	1.60	.		
	Total	77	0.00	94.70	11.90	25.36		
CD10	AML M0	11	0.00	0.10	0.01	0.03	0.429	0.827
	AML M1	21	0.00	3.20	0.35	0.94		
	AML M2	20	0.00	97.00	5.13	21.65		
	AML M3	8	0.00	2.00	0.36	0.73		

	AML M4	2	0.00	0.00	0.00	0.00		
	AML M6	2	0.00	0.00	0.00	0.00		
	AML M7	0	.	.	.	.		
	Total	64	0.00	97.00	1.76	12.12		
CD19	AML M0	10	0.00	72.00	28.41	36.04	0.989	0.431
	AML M1	22	0.00	77.00	19.09	25.35		
	AML M2	23	0.00	88.70	24.57	33.56		
	AML M3	8	0.00	63.00	8.69	21.99		
	AML M4	2	0.00	50.00	25.00	35.36		
	AML M6	5	0.00	0.50	0.30	0.27		
	AML M7	0	.	.	.	.		
	Total	70	0.00	88.70	19.86	29.20		
CD117	AML M0	10	0.00	87.00	45.84	34.01	2.766	0.018 (Sig)
	AML M1	23	0.00	99.00	61.37	29.40		
	AML M2	27	0.00	94.30	59.18	25.67		
	AML M3	9	0.00	82.00	30.17	34.02		
	AML M4	4	0.00	59.80	23.45	29.05		
	AML M6	6	54.00	97.00	72.47	17.32		
	AML M7	1	45.40	45.40	45.40	.		
	Total	80	0.00	99.00	53.92	30.49		
CD34	AML M0	11	48.00	94.00	74.06	14.55	5.744	<0.001 (Sig.)
	AML M1	22	0.00	98.00	66.32	34.64		
	AML M2	24	0.00	99.00	69.43	34.25		
	AML M3	10	0.00	53.00	10.45	22.03		
	AML M4	4	0.00	86.00	50.98	36.38		
	AML M6	6	0.00	96.00	48.00	39.89		
	AML M7	1	0.00	0.00	0.00	.		
	Total	78	0.00	99.00	58.16	36.90		
CD16	AML M0	0	.	.	.	.	2.039	0.254
	AML M1	1	0.30	0.30	0.30	.		
	AML M2	0	.	.	.	.		
	AML M3	1	0.10	0.10	0.10	.		
	AML M4	2	1.50	6.60	4.05	3.61		
	AML M6	4	0.70	4.10	2.95	1.53		
	AML M7	1	7.80	7.80	7.80	.		
	Total	9	0.10	7.80	3.12	2.76		

CD14	AML M0	0	.	.	.	.	0.973	0.494
	AML M1	1	14.20	14.20	14.20	.		
	AML M2	2	0.10	3.00	1.55	2.05		
	AML M3	1	0.20	0.20	0.20	.		
	AML M4	4	12.70	87.00	35.45	34.62		
	AML M6	4	6.10	11.20	7.58	2.44		
	AML M7	1	8.50	8.50	8.50	.		
	Total	13	0.10	23.00	9.55	7.08		
CD64	AML M0	1	14.20	14.20	14.20	.	0.668	0.663
	AML M1	1	3.90	3.90	3.90	.		
	AML M2	2	4.10	19.00	11.55	10.54		
	AML M3	0	.	.	.	.		
	AML M4	3	21.80	93.00	46.37	40.40		
	AML M6	4	16.70	61.00	28.88	21.45		
	AML M7	1	7.80	7.80	7.80	.		
	Total	12	3.90	93.00	25.30	25.94		
CD36	AML M0	1	6.00	6.00	6.00	.	19.140	0.001
	AML M1	0	.	.	.	.		
	AML M2	0	.	.	.	.		
	AML M3	1	0.30	0.30	0.30	.		
	AML M4	4	19.00	27.00	24.43	3.66		
	AML M6	4	19.00	24.90	22.53	2.52		
	AML M7	1	10.50	10.50	10.50	.		
	Total	11	0.30	27.00	18.60	9.03		
CD11b	AML M0	0	.	.	.	.	17.223	0.025
	AML M1	0	.	.	.	.		
	AML M2	0	.	.	.	.		
	AML M3	0	.	.	.	.		
	AML M4	3	21.00	41.30	31.43	10.16		
	AML M6	2	0.00	0.00	0.00	0.00		
	AML M7	0	.	.	.	.		
	Total	5	0.00	41.30	18.86	18.66		
CD15	AML M0	0	.	.	.	.	–	–
	AML M1	0	.	.	.	.		
	AML M2	0	.	.	.	.		
	AML M3	1	34.00	34.00	34.00	.		

AML M4	1	71.00	71.00	71.00	.
AML M6	0	.	.	.	.
AML M7	0	.	.	.	.
Total	2	34.00	71.00	52.50	26.16

Mean expression of HLADR of patients with different morphological diagnosis (n=53) was found to be statistically significant (p=0.001). HLADR expression was found to be lower in patients diagnosed as AML-M3 (9.90±20.98%) and as AML-M4 (44.50±30.47%) as compared to patients with other morphological diagnoses.

Mean expression of MPO of patients with different morphological diagnosis (n=61) was found to be statistically significant (p=0.002). MPO expression was found to be 0.00% in patients diagnosed as AML-M7 and was lower in patients diagnosed as AML-M6 (14.18± 34.74%), AML-M4 (32.67± 33.01%) and AML-M0 (36.88± 32.45%) as compared to patients with other morphological diagnoses.

Mean expression of CD117 of patients with different morphological diagnosis (n=80) was found to be statistically significant (p=0.018). CD117 expression was found to be lower in patients diagnosed as AML-M4 (23.45±29.05%) and as AML-M3 (30.17±34.02%), AML-M7 (45.40%), AML-M0 (45.84±34.01%) as compared to patients with other morphological diagnosis.

Mean expression of CD34 of patients with different morphological diagnosis (n=78) was found to be statistically significant (p<0.001). CD34 expression in patients diagnosed as AML-M7 was found to be 0.00% , was found to be lower in patients diagnosed as AML-M3 (10.45±22.03%) as compared to patients with other morphological diagnosis.

Mean expression of CD14 of patients with different morphological diagnosis (n=13) was not found to be statistically significant (p=0.494). CD14 expression was found to be lower in patients diagnosed as AML-M3 (0.20%), AML-M2 (1.55±2.05%) as compared to AML-M4 (35.45±34.62%), AML-M1 (14.20%), AML-M7 (8.505), AML-M6 (7.58±2.54%).

Mean expression of CD36 of patients with different morphological diagnosis (n=11) was found to be statistically

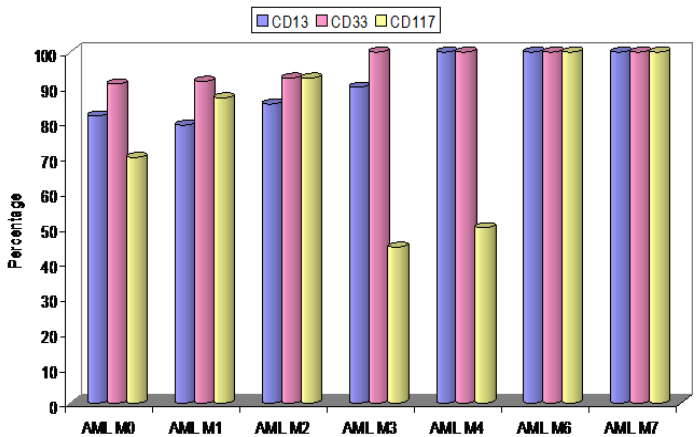
significant (p=0.001). CD36 expression was found to be lower in patients diagnosed as AML-M3 (0.30%), AML-M0 (6.00%) as compared to patients with other morphological diagnosis.

Mean expression of CD11b was done in 5 patients, expression in 2 patients diagnosed as AML-M6 was 0.00% while in patients diagnosed as AML-M4 was found to be 31.43+10.16%. Difference in CD11b expression in patients diagnosed as AML-M6 and AML-M4 was found to be statistically significant (p=0.025).

Table 11a: Comparison of Immune Markers positivity in different morphological diagnosis

	M0	M1	M2	M3	M4	M6	M7	Tota	²	P
CD13										
<20%	2	5	4	1	0	0	0	12	2.935	0.817
	18.18	20.83	14.81	10.00	0.00	0.00	0.00	14.46		
>20%	9	19	23	9	4	6	1	71		
	81.82	79.17	85.19	90.00	100.00	100.00	100.00	85.54		
Total	11	24	27	10	4	6	1	83		
CD33										
<20%	1	2	2	0	0	0	0	5	1.846	0.933
	9.09	8.33	7.41	0.00	0.00	0.00	0.00	6.02		
>20%	10	22	25	10	4	6	1	78		
	90.91	91.67	92.59	100.00	100.00	100.00	100.00	93.98		
Total	11	24	27	10	4	6	1	83		
CD117										
<20%	3	3	2	5	2	0	0	15		

15.785	0.015							
	30.00	13.04	7.41	55.56	50.00	0.00	0.00	
18.75								
>20%	7	20	25	4	2	6	1	65
	70.00	86.96	92.59	44.44	50.00	100.00	100.00	
81.25								
Total	10	23	27	9	4	6	1	80



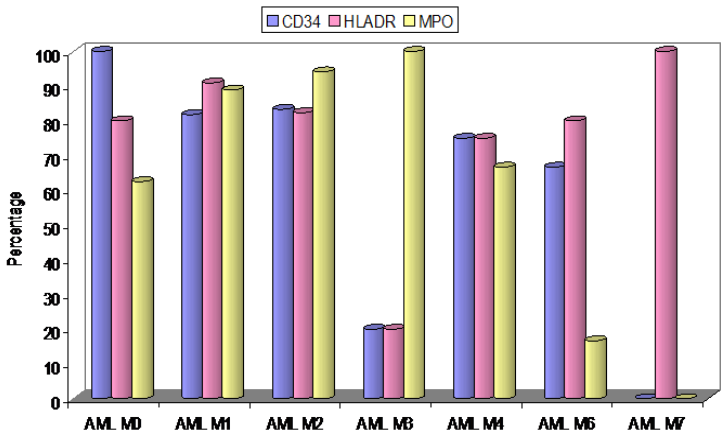
The above table shows that expression of marker CD13 was seen in 85.54% of cases (71/83). Maximum positivity was seen in M4,M6 and M7 (100% cases) of AML followed by AML M3, M2, M0 and M1.

The above table shows that expression of marker CD33 was seen in 93.98% of cases (78/83). Maximum positivity was seen in M3,M4,M6 and M7 (100% cases) of AML followed by AML M2, M1 and M0.

The above table shows that expression of marker CD117 was seen in 81.25% of cases (65/80). Maximum positivity was seen in M6 and M7 (100% cases) of AML followed by AML M2, M1, M0, M4 and M3. These findings came out to be significant.

Table 11b: Comparison of Immune Markers positivity in different morphological diagnoses

	M0	M1	M2	M3	M4	M6	M7	Total
^{2}	P							
HLADR								
<20%	1	1	3	8	1	1	0	15
16.879	0.010							
	20.00	9.09	17.65	80.00	25.00	20.00	0.00	28.30
>20%	4	10	14	2	3	4	1	38
	80.00	90.91	82.35	20.00	75.00	80.00	100.00	71.70
Total	5	11	17	10	4	5	1	53
MPO								
<10%	3	2	1	0	1	5	1	13
24.661	0.000							
	37.50	11.11	5.88	0.00	33.33	83.33	100.00	21.31
>10%	5	16	16	8	2	1	0	48
	62.50	88.89	94.12	100.00	66.67	16.67	0.00	78.69
Total	8	18	17	8	3	6	1	61
CD34								
<20%	0	4	4	8	1	2	1	20
24.034	0.001							
	0.00	18.18	16.67	80.00	25.00	33.33	100.00	25.64
>20%	11	18	20	2	3	4	0	58
	100.00	81.82	83.33	20.00	75.00	66.67	0.00	74.36
Total	11	22	24	10	4	6	1	78



The above table shows that expression of marker CD34 was seen in 74.36% of cases (58/78). Maximum positivity was seen in M0 (100% cases) of AML followed by AML M2 and M1.Least positivity was seen in AML M7 and AML M3

The above table shows that expression of marker HLADR was seen in 71.70% of cases (38/53). Maximum positivity was seen in M7,M1,M2 types of AML. Least positivity was seen in AML M3.

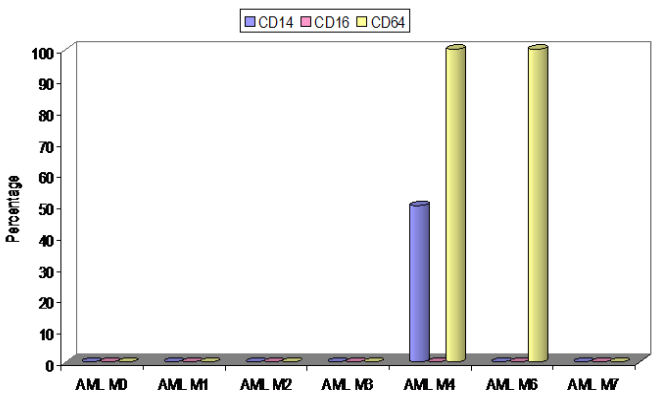
The above table shows that expression of marker MPO was seen in 78.69% of cases (48/61). Maximum positivity was seen in M3 (100% cases), followed by AML M2 and M1. Least positivity was seen in AML M7.

Minimal expression of CD34 and HLADR in AML M3 was found to be statistically significant.

Table 11c: Comparison of Immune Markers positivity in different morphological diagnosis

	M0	M1	M2	M3	M4	M6	M7	Total	^{2}	P
CD14										
<20%	–	1	2	1	2	4	1	11	5.318	0.378
	–		100.00	100.00		100.00	50.00	100.00	100.00	84.62
>20%	–		0	0		0	2	0	0	2

	–	0.00	0.00	0.00	50.00	0.00	0.00	15.38
Total	–	1	2	1	4	4	1	13
CD16								
<20%	–	1	–	1	2	4	1	9
–	–							
	–	100.00	–	100.00	100.00	100.00	100.00	100.00
>20%	–	0	–	0	0	0	0	0
	–	0.00	–	0.00	0.00	0.00	0.00	0.00
Total	–	1	–	1	2	4	1	9
CD64								
<20%	1	1	2	–	0	3	1	8
8.625	0.125							
	100.00	100.00	100.00	–	0.00	75.00	100.00	66.67
>20%	0	0	0	–	3	1	0	4
	0.00	0.00	0.00	–	100.00	100.00	0.00	33.33
Total	1	1	2	–	3	4	1	12



The above table shows that expression of marker CD14 was seen in 15.38% of cases (2/11). Positivity was seen in M4 (50% cases) while all the other subtypes did not show its expression.

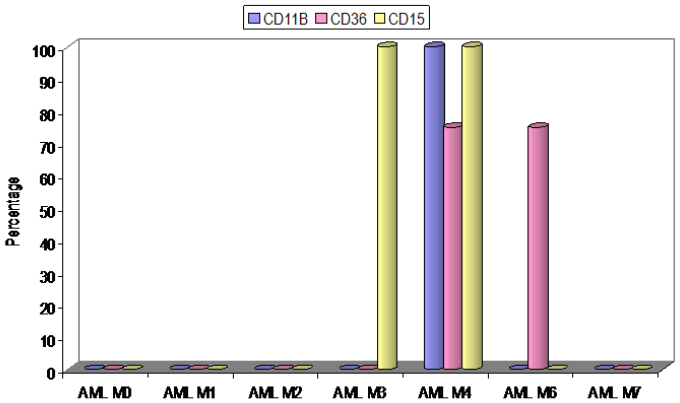
The above table shows that expression of marker CD16 was not seen in any of the 18 cases in which it was interpreted.

Also, the table shows that expression of marker CD64 was seen in 33.33% of cases (4/12). Maximum positivity was seen in M4 and M6 (100% cases) and no expression was seen in other types.

Table 11d: Comparison of Immune Markers positivity in different morphological diagnoses

	M0	M1	M2	M3	M4	M6	M7	Total	^{2}	P
CD36										
<20%	1	–	–	1	1	1	1	5	4.950	0.292
	100.00	–	–		100.00	25.00	25.00	100.00		45.45
>20%	0	–	–		0	3	3	0		6
	0.00	–	–		0.00	75.00	75.00	0.00		54.55
Total	1	–	–		1	4	4	1		11
CD11b										
<20%	–	–	–		–	0	2	–		2
5.000	0.025									
	–	–	–		–	0.00	100.00	–		40.00
>20%	–	–	–		–	3	0	–		3
	–	–	–		–	100.00	0.00	–		60.00
Total	–	–	–		–	3	2	–		5
CD15										
<20%	–	–	–		0	0	–	–		

–	–						
	–	–	–	0.00	0.00	–	–
>20%	–	–	–	1	1	–	–
	–	–	–	100.00	100.00	–	–
Total	–	–	–	1	1	–	–



The table below shows that the expression of marker CD36 was seen in 54.55% of cases (6/11). Positivity was seen in M4 and M6 (75% cases) while all the other subtypes did not show its expression.

The table below shows that the expression of marker CD11b was seen in 60% of cases (3/5). Positivity was seen in M4 (100% cases) while all the other subtypes did not show its expression.

The table below shows that the expression of marker CD15 was seen in AML M3 and M4 cases.

Findings related to expression of immune markers HLADR, MPO, CD117, CD34, CD11B were found to be statistically significant.

Table 11e: Comparison of Immune Markers positivity in different morphological diagnoses

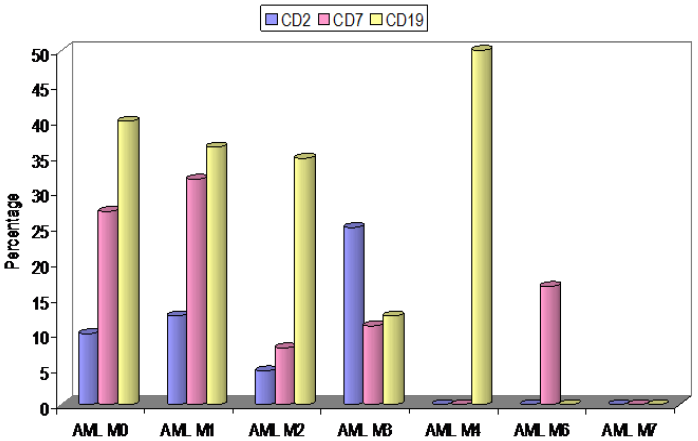
	M0	M1	M2	M3	M4	M6	M7	Total	²	P
CD3										
<20%	5	12	11	5	2	–	–	35		

–	–							
	100.00	100.00	100.00	100.00	100.00	–	–	
100.00								
Total	5	12	11	5	2	–	–	35
CD10								
<20%	11	21	19	8	2	2	–	63
2.235	0.816							
	100.00	100.00	95.00	100.00	100.00	100.00	–	
98.44								
>20%	0	0	1	0	0	0	–	1
	0.00	0.00	5.00	0.00	0.00	0.00	–	
1.56								
Total	11	21	20	8	2	2	–	64
CD11c								
<20%	–	–	–	–	0	–	–	0
–	–							
	–	–	–	–	0.00	–	–	
0.00								
>20%	–	–	–	–	1	–	–	1
	–	–	–	–	100.00	–	–	
100.00								
Total	–	–	–	–	1	–	–	
1								

CD10 was found to be positive in only 5% cases of AML M2 and CD11c in 100% cases of AML M4while CD3 was found in none of the subtypes.

Table 12: Aberrant expression of immune markers in different AML subtypes.

	M0	M1	M2	M3	M4	M6	M7	Total	χ^2	P
CD2										
<20%	9	14	20	3	2	2	–	50	2.336	0.801
	90.00	87.50	95.24	75.00	100.00	100.00	–			90.91
>20%	1	2	1	1	0	0	–	5		
	10.00	12.50	4.76	25.00	0.00	0.00	–			9.09
Total	10	16	21	4	2	2	–	55		
CD7										
<20%	8	15	23	8	3	5	1	63		
6.304	0.390									
	72.73	68.18	92.00	88.89	100.00	83.33	100.00			81.82
>20%	3	7	2	1	0	1	0	14		
	27.27	31.82	8.00	11.11	0.00	16.67	0.00			18.18
Total	11	22	25	9	3	6	1	77		
CD19										
<20%	6	14	15	7	1	5	–	48		
4.651	0.460									
	60.00	63.64	65.22	87.50	50.00	100.00	–			68.57
>20%	4	8	8	1	1	0	–	22		
	40.00	36.36	34.78	12.50	50.00	0.00	–			31.43
Total	10	22	23	8	2	5	–	70		



The following table shows that aberrant expression of marker CD2 was present in 9.09% (5 out of 55 cases) of AML. Maximum positivity was seen in AML M3 (25% cases) followed by AML M1, M0 and M2 and absent in AML M4, M6 and M7.

The table also shows that aberrant expression of marker CD7 was present in 18.18% (14 out of 77 cases) of AML.Maximum positivity was seen in AML M1 (31.82% cases) followed by AML M0, M6 and M2 and absent in AML M4 and M7.

We can also infer from the following table that the aberrant expression of marker CD19 was present in 31.43% (22 out of 70 cases) of AML. Maximum positivity was seen in AML M4 (50% cases) followed by AML M0, M1,M2 and M3 and absent in AML M6 and M7.

Table 13: Comparison of cytogenetics found in different morphological diagnoses

	M0	M1	M2	M3	M4	M6	M7	Total	^{2}	P
t(8;21)										
Negative	5	8	5	7	1	4	30	12.593	0.028	
	100.00	100.00	55.56	100.00	100.00	100.00	88.24			
Positive	0	0	4	0	0	0				4

	0.00	0.00	44.44	0.00	0.00	0.00		11.76
Total	5	8	9	7	1	4		34
1(16;16)								
Negative	5	8	9	7	1	4	–	34
–	–							
	100.00	100.00	100.00	100.00	100.00	100.00	–	100.00
Positive	0	0	0	0	0	0	–	0
	0.00	0.00	0.00	0.00	0.00	0.00	–	0.00
Total	5	8	9	7	1	4	–	34
t(15,17)								
Negative	5	8	9	0	1	4	–	27
34.000	<0.001							
	100.00	100.00	100.00	0.00	100.00	100.00	–	79.41
Positive	0	0	0	7	0	0	–	7
	0.00	0.00	0.00	100.00	0.00	0.00	–	20.59
Total	5	8	9	7	1	4	–	34
Deletion 20/20q								
Negative	–	0	–	–	–	–	–	0
–	–							
	–	0.00	–	–	–	–	–	0.00
Positive	–	1	–	–	–	–	–	1
	–	100.00	–	–	–	–	–	100.00
Total	–	1	–	–	–	–	–	1

Cytogenetics was done in only 35 cases out of 95. Out of three translocations, t (15;17) was found to be the most common. It was detected in 20.59% (7/34) of cases.Infact, all the cases (100%) of

AML M3 were positive for this translocation in which it was done. All the other subtypes of AML were negative for t(15;17) in the cases in which it was performed.

The translocation t(8;21) was found to be the next most common.It was detected in 11.76% (4/34) of cases.Infact, 4 out of 9 cases (44.44%) of AML M2 were positive for this translocation. All the other subtypes of AML were negative for t(8;21) in the cases in which it was performed. These findings were found to be statistically significant.

The translocation t(16;16) was not detected in any of the AML subtypes, in the cases in which it was done.